# *Bemisia tabaci* (Homoptera: Aleyrodidae) Biotype B Colonization on Okra- and Normal-Leaf Upland Cotton Strains and Cultivars

CHANG-CHI CHU,[1] ERIC T. NATWICK,[2] AND THOMAS J. HENNEBERRY[2]

Western Cotton Research Laboratory, USDA-ARS, 4135 E. Broadway Road, Phoenix, AZ 85040

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**ABSTRACT** We compared smooth-leaf okra- and normal-leaf upland cotton (*Gossypium hirsutum* L.) strains and cultivars for susceptibility to colonization by *Bemisia tabaci* (Gennadius) biotype B. Experiments were conducted at seven field sites, five at Holtville, CA, and two at Maricopa, AZ, during 1996–2000. Okra-leaf strains and cultivars, as a group, had lower numbers of adults, eggs, and nymphs compared with normal-leaf strains and cultivars indicating the potential of okra-leaf genetic traits for reducing colonization by *B. tabaci.* Results also suggest that okra-leaf shape may provide less favorable micro-environmental conditions for the habitat of *B. tabaci* because of more open canopy as evidenced by higher leaf perimeter to leaf area ratio. The okra-leaf cultivar 'Siokra L-23' appears to have genetic traits that should be examined further as a source of *B. tabaci* resistance.

**KEY WORDS** *Bemisia tabaci* biotype B, *Bemisia argentifolii,* okra-leaf, normal-leaf, cotton, host plant resistance

DESPITE EXTENSIVE EFFORTS to control *Bemisia tabaci* (Gennadius) biotype B in cotton, 341,541 ha were found infested with the pest in the United States in 2000 (William 2001). A long-term *B. tabaci* management solution that is both economical and environmentally acceptable is needed. In the United States, differences in cotton plant susceptibility to colonization by *B. tabaci* biotype B, also referred to as *B. argentifolii* Bellows & Perring, were reported between upland, *Gossypium hirsutum* L., and Pima, *G. barbadense* L., cottons (Natwick et al. 1995, Percy et al. 1997) and among different upland cottons cultivars (Chu et al. 1998, 1999). These differences were attributed to variations in leaf pubescence—genotypes with more trichomes are generally more susceptible to *Bemisia* infestation than genotypes with smooth leaf characteristics (Butler and Henneberry 1984, Flint and Parks 1990, Butler et al. 1991, Norman and Sparks 1997, Percy et al. 1997), and to differences in leaf shapes—okra-leaf genotypes are generally more resistant to *Bemisia* than normal-leaf genotypes (Berlinger 1986, Chu et al. 1999). Normal- and okra-leaf are defined as the cotton leaf shape that has leaf lobes with shallow and deep indention between lobes, respectively.

The development of insect resistant cultivars by conventional plant breeding and selection are approaches that warrant increased attention for insect population suppression (Sippell et al. 1987). Where successful host plant resistance was developed, the adverse effect on insect populations were dramatic (Wiseman 1999). The methodology is readily accepted as economically sound (Jenkins 1999). We have conducted a number of studies to identify mechanisms of cotton plant resistance to *B. tabaci* colonization (Chu et al. 1998, 1999, 2000, 2001; Freeman et al. 2001). The objective of the current study was to identify cotton leaf shape as a potential genetic trait for the breeding of cottons resistant to *Bemisia.*

## Materials and Methods

Two field experiments were conducted in 1999 and 2000 at the University of Arizona Maricopa Agricultural Research Center at Maricopa, AZ. Five field experiments were conducted from 1996 to 2000 at the University of California Desert Research and Extension Center at Holtville, CA. The soil at Maricopa is Casa Grande sandy loam and the soil at Holtville is Holtville silty clay (Perrier et al. 1974, Post et al. 1988). The experimental design for all experiments was a randomized complete block with four replicates. Thirty-seven smooth okra- and normal-leaf cotton strains and cultivars were compared (Table 1). Both okra- and normal-leaf strains and cultivars were upland cottons having few leaf trichomes, ≈20/cm$^2$ disk or less branched stellate trichomes on the underleaf surface of fifth node main stem leaves as compared with 120/cm$^2$ disk for hairy leaf cultivar such as 'Stoneville 474' (Chu et al. 2001). Not all strains and cultivars were tested in each experiment because of the availability of a seed source (e.g., 87031–26), and resources to

[1] E-mail: cchu@wcrl.ars.usda.gov.

[2] University of California, Desert Research and Extension Center, Holtville, CA 92250.

**Table 1. Smooth okra-and normal-leaf cotton strains and cultivars studied for susceptibility to colonization by *Bemisia tabaci* at Maricopa, AZ, and Holtville, CA, 1996 to 2000**

| Strains/cultivars[a] | Leaf type O or N[b] | Holtville | | | | | Maricopa | |
|---|---|---|---|---|---|---|---|---|
| | | 1996 | 1997 | 1998 | 1999 | 2000 | 1999 | 2000 |
| 87031-126 | O | X[c] | | | | | | |
| 89013-114 | O | X | | | | | X | |
| 89230-244-1028 | O | | | X | X | | | |
| 89230-341-907 | O | | X | | | | | |
| 91209-194 | O | | | | X | | | |
| 91212-265 | O | | | X | | | | |
| E0223 | O | | | | | X | | X |
| E0798 | O | | | | | X | | X |
| E1028 | O | | | | | X | | X |
| FiberMax 819 | O | | | X | X | X | X | |
| FiberMax 832 | O | | | X | X | X | X | |
| FiberMax 975 | O | | | X | | | | |
| Siokra I-4 | O | | X | | | | X | |
| Siokra I-4/649 | O | X | | | | | | |
| Siokra L23 | O | X | X | X | X | X | X | X |
| Siokra S-101 | O | | X | | | | | |
| Siokra V-15 | O | X | X | | | | | |
| C118-293 | N | | X | | | | | |
| CS 50 | N | X | X | | | | | |
| DP 20 | N | X | X | X | X | X | | |
| DP 50 | N | X | X | X | X | X | | |
| DP 90 | N | X | X | X | X | X | | |
| DP 20B | N | | | | | | X | X |
| DP 50B | N | | | | | | X | X |
| DP 90B | N | | | | | | X | X |
| DP 5415 | N | X | X | X | X | X | | |
| DP 5432 | N | X | X | X | X | X | | |
| DP 5461 | N | X | X | X | X | X | | |
| DP 5557 | N | | | X | X | X | | |
| DP 9050 | N | X | | | | | | |
| DP 9057 | N | X | X | | | | | |
| DPX 9775 | N | | | X | | | | |
| HCR 7126 | N | | | | X | | | |
| HCR 9240 | N | | | | X | X | | |
| HCR 9257 | N | | | | X | X | | |
| NuCOTN 33B | N | X | | | | | X | X |
| Texas 121 | N | | X | X | | | | |
| Total | O | 5 | 5 | 6 | 5 | 6 | 5 | 4 |
| | N | 10 | 10 | 9 | 10 | 9 | 4 | 4 |

[a] Cotton seeds were provided for Siokra cultivars, C118-293, and CS 50 by Cotton Seed Distributors Ltd., Dalby, Queensland, Australia; numbered (okra-leaf) strains including the three E strains by Australia's Commonwealth Scientific and Industrial Research Organization, Narrabri, Australia; the three FiberMax cultivars by Aventis CropScience, Research Triangle Park, NC; all DP, DPX, and NuCOTN 33B by Delta and Pine Land Co, Scott, MS; the three HCR cultivars by Helena Cotton Research, Casa Grande, AZ; and Texas 121 by Syngenta Seed, Victoria, TX.

[b] O and N denote smooth okra- and normal-leaf strains or cultivars, respectively.

[c] X denotes the strains and cultivars tested in experiment.

manage the test sites. Standard agronomic practices were followed at each test site.

Plots at Maricopa, AZ, were eight rows wide and 12.2 m long with rows 1 m apart. There were two unplanted rows between plots and 3-m wide alleys between blocks. Seeds were planted and watered for germination on 19 April for 1999 and 13 April for 2000. Plants emerged ≈2 wk later and were watered at 10–20 d intervals during their growing seasons. Plots were not treated with any insecticides except with diflubenzuron (1-[4-chlorophenyl-3-(2,6-difluorobenzoyl urea)]) for the control of saltmarsh caterpillars, *Estigmene acrea* (Drury) (Lepidoptera: Arctiidae) on 13 August in 1999.

Plots at Holtville, CA, were four rows wide except in 1999 when plots were eight rows wide. Rows were 14–15 m long and 1 m apart between rows. There were unplanted rows between plots and 4–5 m wide valleys between blocks. Seeds were planted and watered for germination on 20, 28, 25, 26, and 24 March each year from 1996 to 2000, respectively.

Densities of *B. tabaci* biotype B on cotton at Maricopa, AZ, were estimated at 7-d intervals from 21 July to 6 October in 1999 and from 10 July to 5 September in 2000. On each sampling date, three plants per plot were randomly selected. Leaves were picked from the first, third, fifth, seventh, 10th, and 15th main stem nodes in 1999 and from the first to fifth and seventh nodes in 2000. The nodes were in ascending numbers beginning with the first expanded leaf below the plant main terminal. Leaves from the first node measured ≥2.5 cm between the two largest leaf lobes. Among the nodes, the 15th was the lowest position on the main stems at the time of sampling. A 2-cm$^2$ leaf disk

**Table 2. Mean ± SE numbers of *Bemisia tabaci* biotype B on smooth okra- and normal-leaf upland cotton strains and cultivars at Maricopa, AZ and Holtville, CA, 1996 to 2000**

| Location | Year | Treatment | $n$ | No. adults/leaf | No./cm² leaf disk | |
|---|---|---|---|---|---|---|
| | | | | | Eggs | Nymphs |
| Maricopa | 1999 | Okra | 5 | 12.0 ± 0.8b | 25.5 ± 1.6a | 10.3 ± 0.9a |
| | | Normal | 4 | 20.9 ± 1.5a | 26.3 ± 2.1a | 10.8 ± 0.9a |
| | 2000 | Okra | 5 | 3.9 ± 0.6b | 5.7 ± 0.8b | 2.5 ± 0.4b |
| | | Normal | 4 | 5.8 ± 0.3a | 12.9 ± 1.0a | 4.8 ± 0.4a |
| Holtville | 1996 | Okra | 5 | 33.5 ± 6.1b | 18.2 ± 2.4b | 12.8 ± 1.4b |
| | | Normal | 10 | 54.2 ± 4.8a | 31.5 ± 2.0a | 23.1 ± 1.4a |
| | 1997 | Okra | 5 | 3.2 ± 0.3b | 1.2 ± 0.2b | 1.0 ± 0.1b |
| | | Normal | 10 | 4.7 ± 0.2a | 2.3 ± 0.1a | 1.6 ± 0.1a |
| | 1998 | Okra | 6 | 5.2 ± 0.2b | 6.9 ± 0.4b | 8.2 ± 0.4b |
| | | Normal | 9 | 6.6 ± 0.2a | 10.0 ± 0.7a | 9.3 ± 0.6a |
| | 1999 | Okra | 5 | 5.6 ± 0.4b | 43.3 ± 5.8b | 43.4 ± 5.4b |
| | | Normal | 10 | 9.8 ± 0.2a | 66.6 ± 2.8a | 62.0 ± 2.6a |
| | 2000 | Okra | 6 | 4.8 ± 0.4b | 21.7 ± 2.1b | 29.0 ± 4.2b |
| | | Normal | 10 | 7.2 ± 0.3a | 30.0 ± 2.2a | 42.6 ± 3.8a |
| Mean | | Okra | | 9.7 ± 4.1b | 17.5 ± 5.5b | 15.3 ± 5.8b |
| | | Normal | | 15.6 ± 6.8a | 25.7 ± 8.0a | 22.0 ± 8.5a |

Means in a column for pair comparison in a year and overall means not followed by the same letters are significantly different by orthogonal comparison ($P = 0.05$).

was taken from the area adjacent to center primary vein at the basal area of the leaves (Naranjo and Flint 1994). Numbers of eggs and nymphs were counted on underleaf disk surfaces with the aid of a stereoscope. Adults per leaf were counted on three fifth main stem node leaves on plants in each plot in 1999 and on three leaves on plants from each of the six main stem nodes sampled in each plot in 2000 using turn-leaf method developed earlier (Naranjo and Flint 1995) on each sampling date. Leaf area and perimeter of each sampled leaf during the growing season were measured with a leaf area meter (CI-400 CIAS Image Analysis, CID, Vancouver, WA).

Densities of *B. tabaci* biotype B on cotton at Holtville, CA, were estimated by picking leaves at 7-d intervals from 17 June to 29 July, from 6 June to 19 August, from 10 June to 12 August, from 30 June to 25 August, and from 29 May to 7 August in 1996–2000, respectively. On each sampling date, fifth main stem node leaves from each of 10 plants in each plot were randomly selected. Egg and nymph numbers were counted from one 1.54- to 1.65-cm² leaf disk punched from each leaf as described earlier. Adults were counted from fifth main stem node leaves of 10 plants.

Data of the multiple *Bemisia* density samplings and leaf area and perimeter measurements of each experiment were first averaged into seasonal means and, then, analyzed using analysis of variance (ANOVA) with means separated with Student-Neuman-Keul's multiple range test (Anonymous 1989). When the treatment effect was significant, an orthogonal comparison was conducted between okra- and normal-leaf cotton strains and cultivars. In addition, an okra-leaf strain or cultivar that consistently showed lower infestation of *Bemisia* was chosen to compare with other okra-leaf strains and cultivars for the potential genetic traits for breeding cultivars resistant to *Bemisia.* The overall mean numbers of *Bemisia* between the okra- and normal- leaf strains and cultivars, and the chosen okra-leaf and other okra-leaf of each experiment were pooled and further analyzed using the seven experiments as the replicates with means separated with *t*-test. Likewise, the overall means of leaf area, perimeter, and the ratio of the two were pooled and further analyzed using the two experiments as the replicates. Data of means were transformed using square root plus one transformation to normalized the heterogeneity before ANOVA when necessary.

## Results

Mean numbers of *B. tabaci* adults on okra-leaf cotton strains and cultivars were significantly lower compared with normal-leaf in 1999 and 2000 at Maricopa, AZ (Table 2; $F = 47.1$ and 16.4 with df = 1, 21, and 24, respectively); and in 1996, 1997, 1998, 1999, and 2000 at Holtville, CA ($F = 12.3$, 130.4, 68.5, 412.4, and 85.5 with df = 1, 42; respectively). Mean numbers of eggs on okra-leaf cotton strains and cultivars were significantly lower compared with normal-leaf in 2000 at Maricopa, AZ ($F = 35.8$ with df = 1, 21); and in 1996, 1997, 1998, 1999, and 2000 at Holtville, CA ($F = 24.8$, 51.3, 39.1, 56.8, 19.3 with df = 1, 42, respectively). Mean numbers of nymphs on okra-leaf cotton strains and cultivars were significantly lower compared with normal-leaf in 2000 at Maricopa, AZ ($F = 20.4$ with df = 1, 21); and in 1996, 1997, 1998, 1999, and 2000 at Holtville, CA ($F = 35.1$, 37.3, 5.3, 55.3, and 12.6 with df = 1, 42, respectively). The exceptions were numbers of eggs and nymphs/cm² of leaf disk in 1999 at Maricopa, AZ, which did not show significant differences between okra- and normal-leaf cotton strains and cultivars ($F = 0.2$ and 0.6 with df = 1, 24). For overall means of the seven experiments, numbers of adults, eggs, and nymphs for okra-leaf were significantly lower compared normal-leaf strains and cultivars ($F = 14.7$, 15.8, and 10.7 with df = 1, 6, respectively).

Mean numbers of adults, eggs, and nymphs for 'Siokra L-23' were not significantly lower compared

**Table 3. Mean ± SE numbers of *Bemisia tabaci* biotype B on Siokra L-23 and other smooth okra-leaf upland cotton strains and cultivars at Holtville, CA and Maricopa, AZ, 1996 to 2000**

| Location | Year | Treatment | *n* | No. adults/ leaf | No./cm$^2$ leaf disk | |
|---|---|---|---|---|---|---|
| | | | | | Eggs | Nymphs |
| Maricopa | 1999 | Siokra L-23 | 1 | 9.1 ± 0.7a | 23.1 ± 5.4a | 8.8 ± 2.2a |
| | | Others | 4 | 12.8 ± 0.9a | 26.1 ± 1.6a | 10.2 ± 0.9a |
| | 2000 | Siokra L-23 | 1 | 2.8 ± 0.3a | 5.4 ± 0.3a | 1.9 ± 0.1a |
| | | Others | 3 | 4.2 ± 0.7a | 5.8 ± 1.0a | 2.7 ± 0.6a |
| Holtville | 1996 | Siokra L23 | 1 | 22.6 ± 11.4a | 11.5 ± 3.8a | 10.3 ± 2.9a |
| | | Others | 4 | 31.1 ± 7.3a | 18.1 ± 3.0a | 12.9 ± 1.8a |
| | 1997 | Siokra L-23 | 1 | 2.0 ± 0.1b | 0.7 ± 0.1b | 0.7 ± 0.1b |
| | | Others | 4 | 3.4 ± 0.4a | 1.4 ± 0.2a | 1.1 ± 0.1a |
| | 1998 | Siokra L-23 | 1 | 4.5 ± 0.2b | 5.1 ± 0.2b | 8.2 ± 1.1a |
| | | Others | 5 | 5.4 ± 0.3a | 7.2 ± 0.5a | 8.2 ± 0.4a |
| | 1999 | Siokra L-23 | 1 | 5.4 ± 0.3a | 37.9 ± 7.4a | 48.0 ± 9.3a |
| | | Others | 4 | 5.6 ± 0.5a | 44.6 ± 7.1a | 42.3 ± 6.4b |
| | 2000 | Siokra L-23 | 1 | 3.9 ± 0.7a | 15.5 ± 4.1b | 20.8 ± 6.6a |
| | | Others | 5 | 5.0 ± 0.4a | 22.9 ± 2.3a | 30.6 ± 4.8a |
| Mean | | Siokra L-23 | 1 | 7.2 ± 2.7b | 14.2 ± 4.9b | 14.1 ± 6.2a |
| | | Others | 6 | 9.6 ± 3.8a | 18.0 ± 5.6a | 15.4 ± 5.8a |

Means in a column for pair comparison in a year and overall means not followed by the same letters are significantly different, orthogonal comparison, ($P$ = 0.05).

with other okra-leaf cotton strains and cultivars at Maricopa, AZ, in 1999 and 2000 (Table 3, $F$ values ranged from 0.6 to 4.1 with df = 1, 12 for 1999 and 1, 9 for 2000). At Holtville, CA, Siokra L-23 was significantly lower than other cotton strains and cultivars in mean numbers of adults in 1997 and 1998 ($F$ = 20.2 and 21.5 with df = 1, 12, and 15, respectively); in mean numbers of eggs in 1997 and 1998 ($F$ = 6.9 and 8.1 with df = 1, 12, and 15, respectively); and in mean numbers of nymphs in 1997 ($F$ = 5.5 with df = 1, 12). Mean numbers of adults, eggs, and nymphs were not significantly different between Siokra L-23 and other okra cotton strains and cultivars in other comparisons in 1996–2000 ($F$ values ranged from 0.0 to 3.6 with df = 1, 12, or 15, respectively). For the overall means of the seven experiments, numbers of adults and eggs of Siokra L-23 were significantly lower compared with other okra-leaf cotton strains and cultivars ($F$ = 16.0 and 11.5 with df = 1, 6). Mean numbers of nymphs/cm$^2$ of leaf disk were not significantly different ($F$ = 0.6 with df = 1, 6).

Mean leaf areas were significantly smaller for okra-leaf cotton strains and cultivars compared with normal-leaf in 1999 (Table 4, $F$ = 20.7 with df = 1, 24) and in 2000 ($F$ = 41.9 with df = 1, 21) at Maricopa, AZ. Mean leaf perimeters were significantly greater for okra-leaf cotton strains and cultivars compared with normal-leaf in 1999 ($F$ = 181.1 with df = 1, 24) and in 2000 ($F$ = 2,161.2 with df = 1, 21). Ratios of leaf perimeters and areas were significantly greater for okra-leaf cotton strains and cultivars compared with normal-leaf in 1991 ($F$ = 190.7 with df = 1, 24) and in 2000 ($F$ = 2,861.6 with df = 1, 21). For the overall means of the two experiments, mean leaf areas were significantly lower for okra-leaf compared with normal-leaf cotton strains and cultivars ($F$ = 4,773.9 with df = 1, 1) and mean leaf perimeters and ratios of leaf perimeter and area were significantly higher for okra-leaf compared with normal-leaf cotton strains and cultivars ($F$ = 171.5 and 2,134.7 with df = 1, 1, respectively).

Whitefly density on the cotton strains and cultivars tested in each experiment varied from one to other, but can be generalized as high and low, respectively, as follows: DP 50B and Siokra L-23 for 1999 and 2000 at Maricopa AZ; DP 9050 and Siokra L-23 for 1996, DP 5461 and Siokra L-23 for 1997, DP 5557 and Siokra L-23 for 1998, DP 90 and Siokra L-23 for 1999, and DP 5432 and Siokra L-23 for 2000 at Holtville, CA.

## Discussion

Our results corroborate results from Sudan that the okra- and super-okra-leaf genotypes confer resistance

**Table 4. Mean ± SE numbers of leaf area (square centimeter) and perimeters (centimeter) of okra- and normal-leaf cotton strains and cultivars at Maricopa, AZ, 1999 to 2000**

| Year | Leaf shape | Leaf area (A) | Leaf perimeter (P) | P/A |
|---|---|---|---|---|
| 1999 | Okra | 49.6 ± 1.5b | 62.1 ± 1.3a | 1.68 ± 0.04a |
| | Normal | 59.0 ± 1.9a | 41.4 ± 0.8b | 0.95 ± 0.03b |
| 2000 | Okra | 55.3 ± 2.1b | 73.4 ± 1.4a | 1.53 ± 0.03a |
| | Normal | 64.9 ± 1.4a | 46.4 ± 0.5b | 0.85 ± 0.02b |
| Mean | Okra | 52.5 ± 2.9b | 67.8 ± 5.7a | 1.61 ± 0.08a |
| | Normal | 62.0 ± 6.0a | 43.9 ± 2.5b | 0.90 ± 0.01b |

Means in a column for pair comparison in a year and overall means not followed by the same letters are significantly different, orthogonal comparison, ($P$ = 0.05).

to *B. tabaci* (Sippell et al. 1987) and okra-leaf cotton genetic traits may have potential for increasing cotton plant resistance to *B. tabaci* infestation. Okra-leaf cultivars have been associated with resistance to a number of other cotton pests (Jenkins 1999). Okra-leaf cotton strains and cultivars have greater leaf perimeters to area ratios compared with normal-leaf strains and cultivars resulting in less shaded area compared with normal-leaf cottons. This leaf characteristic may result in higher ambient temperature and lower humidity in the cotton canopy. Wilson (1994) reported that okra-leaf resistance to the twospotted spider mite, *Tetranychus urticae* Koch, in Australia had been attributed to a reduction in availability of protected sites for oviposition on the underleaf surfaces of leaves. Protected oviposition sites are important because mite eggs are susceptible to high mortality from desiccation in high temperature and low humidity conditions in Australia's cotton producing region and female mites prefer to feed and oviposit on protected leaf surfaces that may have higher boundary layer humidity. We (Chu et al. 1995) reported earlier that *B. tabaci* feed and oviposit almost exclusively on underleaf surfaces. A similar humidity relationship for *B. tabaci* could partially explain the resistance of some okra-leaf genotypes. However, in the United States, nymph (Flint and Parks 1990) or adult (Butler and Wilson 1984) densities on the okra-leaf WC-12NL were not significantly different compared with normal-leaf DP 61. These results imply that *B. tabaci* resistance in okra-leaf cotton is complex and may be influenced by factors other than leaf shape. Other characteristics suggested from earlier studies are distance from the underleaf surface to the center of nearest minor vascular bundles (Cohen et al. 1996) and leaf hairiness (Flint and Parks 1990, Norman and Sparks 1997). Leaf hairiness is associated with increased boundary layer humidity on leaf surfaces (Burrage 1971). In the desert southwestern United States, with extremely high air temperatures and low relative humidity and limited irrigation, a subtle change in underleaf surface humidity could influence *B. tabaci* egg and nymph survival (Chu et al. 2001).

Among the 17 okra-leaf cultivars in our study, Siokra L-23 appears to support lower *B. tabaci* infestation compared with other okra-leaf cultivars.

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